

Comparison of Three Methods for Recovery of Yeasts from Hands of Health-Care Workers

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This study compared three methods for the detection of yeasts on the hands of 30 nurses: (i) direct finger impressions on inhibitory mold agar plates, (ii) bag washes in brain heart infusion broth, and (iii) bag washes in brain heart infusion broth supplemented with gentamicin and vancomycin. The antimicrobial agent-supplemented bag wash method identified the greatest number of yeast carriers and yielded the most yeast isolates, especially non-*C. albicans* *Candida* spp.

In an earlier study, we found that 75% of the nurses in our Medical Center harbored yeasts on their hands (10), a result that contrasted sharply with those from previous studies (1, 6, 9). We hypothesized that the discrepancy between our recovery rate and those reported in the literature was due, at least in part, to differences in methodology. Accordingly, in the present study, we compared three methods for the detection of yeasts on the hands of health-care workers: (i) direct finger impressions on agar plates with fungal medium, (ii) bag wash cultures in broth without antimicrobial agent supplementation, and (iii) bag wash cultures in broth with antimicrobial agent supplementation.

Thirty nurses participated in this study. At the time of the study, 10 nurses were working in the nursing home care unit, 10 were working in intensive care units, and 10 were working in the outpatient clinics. Each nurse was studied in the course of a normal work shift without any mandated hand washing or other specific preparation for participation in the study.

Direct finger impressions on inhibitory mold agar plates were obtained first. The second and third phalanx of fingers from both hands were pressed onto the surface of the agar plates. Next, the bag washes in brain heart infusion broth were performed with standard bag broth techniques (4, 5). Half of the nurses placed their dominant hand into a polyethylene bag containing 20 ml of brain heart infusion broth, and the other half placed their nondominant hand into this medium. Similarly, in the next stage of the sampling, half of the nurses placed their dominant hand in brain heart infusion broth supplemented in advance with gentamicin and vancomycin in concentrations of 50 µg/ml each, and the other half placed their nondominant hand in this supplemented medium. After the wash was performed, broth specimens were poured into sterile plastic cups and transported to the laboratory, where they were processed within 2 h of collection.

When the cups were received in the laboratory, an aliquot (0.1 ml) was removed for subculture to inhibitory mold agar and bromocresol green agar (PML Microbiologicals, Tualatin,

Oreg.). Cups with the remaining 19.9-ml broth specimens and all agar plates were incubated at 30°C for 6 days. Additional aliquots were removed from the cups and subcultured on the two types of agar media when the broth media became turbid and on day 6 of incubation regardless of turbidity status. Control cultures of medium-containing bags (no hand washing and no antibiotics) were performed simultaneously. No yeasts were recovered from the control cultures. Yeasts recovered were identified with MicroScan Rapid Yeast Identification panels (Baxter Healthcare Corporation, West Sacramento, Calif.) (3). Several tests of statistical significance were used to evaluate the data collected in this study (8). All probability values were established on the basis of two-tailed analyses.

The antimicrobial agent-supplemented bag wash method was the most sensitive method for detecting yeast carriage. Overall, 24 (80%) of the 30 nurses for whom samples were cultured yielded a yeast isolate by one technique or another (Table 1). The percentages of the 24 positive nurses detected were 29% with the direct impression method, 62% with the basic bag wash method, and 83% with the antimicrobial agent-supplemented bag wash method. The overall difference between these recovery rates was highly significant; pairwise comparisons of the recovery rates with the three techniques indicated that both bag wash procedures were superior to the direct impression method but not to each other.

The supplemented bag wash technique also yielded the greatest number of yeast isolates from the hands of nurse participants. Overall, 45 different yeasts were recovered by one technique or another. The percentages of the 45 yeasts recovered were 22% with the direct impression method, 47% with the basic bag wash method, and 64% with the supplemented bag wash method. The overall difference between the three methods for recovery of different yeasts from the study nurses was statistically significant. Pairwise comparisons of the numbers of different yeasts recovered by the three different techniques indicated that the supplemented bag wash method recovered significantly more yeasts than the direct impression method. The number of different yeasts detected with the basic bag wash procedure did not differ significantly from those recovered with the other two methods. The yeasts recovered by the three methods encompassed seven genera (Table 2). Of note, 22 *Candida* isolates were recovered with the supplemented bag wash technique. In contrast, only two and nine *Candida* isolates were recovered with the direct impression and

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TABLE 1. Comparison of three culture methods for detection of yeasts on hands of nurses^a

| Nurse group (n) | No. of carriers detected (no. of different isolates recovered) by: | | | |
|-----------------------------|--|----------------------|-----------------------|------------|
| | Direct impression | Basic bag wash | Supplemented bag wash | Any method |
| Nursing home care unit (10) | 4 (7) | 6 (8) | 8 (11) | 10 (19) |
| Intensive care unit (10) | 2 (2) | 4 (5) | 5 (8) | 6 (11) |
| Outpatient clinic (10) | 1 (1) | 5 (8) | 7 (10) | 8 (15) |
| Total (30) | 7 (10) ^b | 15 (21) ^b | 20 (29) ^b | 24 (45) |

^a In Cochran's *Q* test, $Q = 13.6$; $0.01 > P > 0.001$ for the difference in numbers of nurse carriers detected by all three sampling methods. In Friedman two-way analysis of variance, $\chi^2 = 8.9$; $P = 0.012$ for the difference in numbers of yeast isolates recovered with all three sampling methods compared.

^b Results for pairwise comparisons of nurse carriers detected with the three sampling methods by McNemar's test for significant changes are as follows: direct impression versus basic bag wash, $\chi^2 = 4.1$, $P < 0.04$; direct impression versus supplemented bag wash, $\chi^2 = 9.6$, $P < 0.002$; basic bag wash versus supplemented bag wash, $\chi^2 = 1.45$, $P = 0.3$. Results for individual comparisons of numbers of different yeasts recovered by the three sampling methods by Wilcoxon's matched-pairs signed-rank test are as follows: direct impression versus basic bag wash, $0.10 > P > 0.05$; direct impression versus supplemented bag wash, $P < 0.01$; basic bag wash versus supplemented bag wash, $P = 0.10$.

basic bag wash techniques, respectively. Inhibitory mold and bromcreosol green agar performed comparably with both the basic bag wash and supplemented bag wash techniques (data not shown). Neither agar medium offered a clear advantage over the other with either of the two bag wash methods.

The results of the present study confirm our previous observations regarding the high frequency of yeast carriage on the hands of nursing personnel. In the former study, 75% of the nurses were found to harbor yeasts on their hands (10). In the present study, yeasts were detected on the hands of 80% of the nurses studied, and *Candida* spp. were recovered from 63% of the nurses studied. The high frequency of yeast carriage observed in these two studies clearly reflects the heightened sensitivity of the bag wash method used for hand sampling in both studies. In the present study, both bag wash methods identified more nurse carriers and more isolates from these carriers than the direct impression method, and the antibiotic-supplemented bag wash consistently tended to recover yeasts more frequently than the basic bag wash.

The combined observations from several other studies have suggested that hand sampling methodology influences the yield of fungal isolates. One study with premoistened swabs to sample the dorsum of the hand and another study with sterile swabs to sample the hands reported yeast recovery rates of 4.5 and 0%, respectively, in healthy adults (6, 9). Using direct finger impressions on the surface of Sabouraud's dextrose agar, Clayton and Noble found that the percentages of subjects colonized with *Candida albicans* were 7.7% in 376 adult patients, 6.8% in 73 pediatric patients, 0.5% in 407 healthy children, and 17% in 23 medical students (1). Other, unspecified yeasts were isolated in 10.6, 0, 1.5, and 0% of the four study groups, respectively. The isolation of *Candida* spp. from 7% of the nurses with the direct impression method in the present study generally parallels the recovery rates reported by Clayton and Noble.

Finally, two studies with bag wash techniques have reported higher fungal recovery rates than studies with swab or direct impression techniques. With a bag wash technique with a phosphate buffer and Tween 80 solution and subsequent subculture onto Sabouraud's dextrose agar with added chloramphenicol,

TABLE 2. Types of yeasts recovered from nurses' hands with three culture methods

| Yeast(s) recovered | No. of yeasts recovered by: | | |
|------------------------------------|-----------------------------|----------------|-----------------------|
| | Direct impression | Basic bag wash | Supplemented bag wash |
| <i>Rhodotorula</i> spp. | 6 | 9 | 5 |
| <i>Candida albicans</i> | 0 | 1 | 0 |
| <i>Candida parapsilosis</i> | 1 | 6 | 12 |
| <i>Candida guilliermondii</i> | 0 | 1 | 5 |
| <i>Candida zeylanoides</i> | 1 | 0 | 2 |
| <i>Candida lusitanae</i> | 0 | 1 | 1 |
| <i>Candida famata</i> | 0 | 0 | 2 |
| <i>Cryptococcus</i> spp. | 2 | 0 | 0 |
| <i>Torulopsis glabrata</i> | 0 | 1 | 0 |
| <i>Torulopsis candida</i> | 0 | 1 | 0 |
| <i>Saccharomyces</i> spp. | 0 | 0 | 1 |
| <i>Sporobolomyces salmonicolor</i> | 0 | 1 | 0 |
| <i>Hansenula anomala</i> | 0 | 0 | 1 |
| Total | 10 | 21 | 29 |

Horn and colleagues detected yeasts on the hands of 80% of 15 nurses working in an oncology unit, 38% of 16 nurses working in a dermatology unit, and 27% of 30 control subjects (2). *Candida parapsilosis* and *Rhodotorula* spp. were the organisms recovered most frequently from these three groups. Similarly, using a bowl wash procedure, Phelps and colleagues detected *C. albicans* on the hands of 26% of the physicians and nurses sampled during the investigation of an outbreak of candidiasis in a special care baby unit (7).

The antibiotic-supplemented bag wash technique may prove useful in epidemiologic studies of nosocomial yeast infections. On the surface, it is reasonable to think that a method which detects more carriers and more yeasts would provide the greatest number of opportunities for detecting cross-infection or cross-colonization in a health-care facility. On the other hand, it is possible that the antibiotic-supplemented bag technique is too sensitive. It may be capable of detecting yeasts sequestered in the cuticle or under the nail that are not readily passed from one individual to another. If this were the case, then many of the yeasts recovered would have no epidemiologic significance. Consequently, at this point, it seems prudent to recommend that the utility of the antibiotic-supplemented bag wash technique in epidemiologic investigations be determined in well-designed studies comparing the clinical significance of its yield with that of other methods for recovering fungi from the hands.

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